Remarks

I. Support for Amendments

The foregoing amendments to the specification have been made to provide proper format for the trademark used in the application in accordance with MPEP § 608.01(v). These amendments to the specification therefore do not add new matter.

Support for the foregoing amendments to the claims may be found throughout the specification. Specifically, support for new claims 65-76 may be found in the specification at pages 8-14, 23, 24, 31-33, 40-47, and throughout the Examples, and in claims 2-13 as originally filed; support for new claims 77-80 may be found in the specification at pages 32-33, 55-56, 62-63, 68-70 and 74-76; support for new claims 81-84 may be found in the specification at pages 62-63, 70-73 and 75-76; support for new claims 85-88 may be found in the specification at pages 56-62; support for the amendments to claims 29 and 50 may be found in the specification at page 23, line 19, and at page 34, lines 22-23 and 27-28; support for the amendments to claim 31 may be found in the specification at pages 8-14, 23, 24, 31-33, 40-47, and in claim 1 as originally filed; support for the amendments to claim 40 may be found in the specification at pages 8-14, 23, 24, 31-33, 40-47; and the amendments to claims 14, 15, 19, 32, 33 and 37-39 have been made at the suggestion of the Examiner in the Office Action. Accordingly, the present amendments do not add new matter, and their entry is respectfully requested.

II. Status of the Claims

By the foregoing amendments, claims 1-13 and 52-64 have been cancelled without prejudice or disclaimer as being drawn to nonelected restriction groups, new claims 65-88 are sought to be entered, and claims 14, 15, 19, 29, 31-33, 37-40 and 50 have been amended. These amendments do not introduce new matter into the application. Upon entry of the foregoing amendments, claims 14-51 and 65-88 are pending in the application, with claims 14, 19, 31 and 40 being the independent claims.

III. Summary of the Office Action

In the Office Action, the Examiner has made one objection to and seven rejections of the claims. Applicants respectfully offer the following remarks to overcome or traverse each of these elements of the Office Action.

IV. The Objection to Claims 14-51

In the Office Action at page 2, third paragraph, the Examiner has objected to claims 14-51 because each of these claims as originally filed encompasses *in vivo* embodiments, while the embodiments elected in Applicants' Reply to Restriction Requirement filed in the present matter on August 21, 2000, are drawn to *in vitro* methods. By the foregoing amendments, claims 14, 15 and 19 (and thus the claims dependent therefrom) have been amended to delete the *in vivo* recitation in each of these claims. In addition, claims 31 and 40 (and thus the claims dependent therefrom) have been amended to insert an analogous *in vitro* recitation. Thus, this objection has been fully accommodated; reconsideration and withdrawal are respectfully requested.

V. The Rejections Under 35 U.S.C. § 112, Second Paragraph

In the Office Action at pages 2-5, the Examiner has rejected claims 14-51 under 35 U.S.C. § 112, second paragraph, as being indefinite. Applicants respectfully traverse this rejection based on the following remarks.

A. The Recitation of "Effective Amount"

In making this rejection, the Examiner first contends that claims 14 and 19 are vague and indefinite for reciting an "effective amount" of a ribosomal protein. *See* Office Action at page 3, second full paragraph. Applicants respectfully disagree with this contention.

Claims 14 and 19 recite the use of an "effective amount" of at least one ribosomal protein in the reaction mixtures used in the claimed methods. The meaning of "effective amount" of a ribosomal protein is clearly described in detail in the specification, particularly at page 33, lines 3-23. In addition, the specification provides detailed protocols for titration assays that enable the ordinarily skilled artisan to determine the optimum concentration of a given ribosomal protein for use in the methods of claims 14 and 19 (*see, e.g.*, Example 1 at pages 71-73, particularly in Tables 3 and 4 at pages 71 and 72, respectively). Hence, based on the clear guidance provided by the present specification, one of ordinary skill could easily determine the amount of a given ribosomal protein that would be effective at driving a recombinational cloning reaction, *i.e.*, what is an "effective amount" of a ribosomal protein as recited in claims 14 and 19.

As the Board has held:

> [35 U.S.C. § 112, second paragraph] merely requires that the claims set forth and circumscribe a particular area with a reasonable degree of precision and particularity. The definiteness of the claim language employed must not be analyzed in a

> vacuum, but always in light of the teachings of the prior art and of the particular application disclosure as it would be interpreted by one having ordinary skill in the pertinent art.

Ex parte Moelands, 3 USPQ2d 1474, 1476 (Bd. Pat. App. Int. 1987) (citing *In re Moore*, 439 F.2d 1232 (CCPA 1971). Since the meaning of the term "effective amount" of a ribosomal protein is made clear from the description contained in the specification, one of ordinary skill would readily understand the meaning of this phrase as used in the present claims. These claims thus comport with the requirements of 35 U.S.C. § 112, second paragraph, as interpreted under *Moelands* and *Moore*. Therefore, Applicants respectfully assert that this portion of the rejection under 35 U.S.C. § 112, second paragraph is in error; reconsideration and withdrawal are therefore respectfully requested.

B. The Recitation of "Substantially Recombine"

The Examiner next contends that claims 14 and 19 are indefinite because the recitation in these claims of "do not substantially recombine" is allegedly unclear. Applicants respectfully disagree with this contention. The phrase "do not substantially recombine" in claims 14 and 19 can be easily understood by the ordinarily skilled artisan, reading this phrase in view of the disclosure in the present specification and information readily available in the art (which, under *Moelands*, must be considered together to determine the definiteness of a claim). Throughout the Examples in the present specification, data are presented indicating the degree to which given sets of recombination sites recombine under certain reaction conditions (*see, e.g.*, data in Tables 3 and 4 at pages 71-72). In addition, the art is replete with information that would permit one of ordinary skill to determine whether or not particular recombination sites will

"substantially recombine" with one another. For example, in Hartley *et al.*, U.S. Patent No. 5,888,732 (Doc. No. AF3, of record; hereinafter "Hartley"), in Examples 1-3 at cols.19-25 (particularly in Tables 3 and 4 at col. 25), data are presented indicating recombination sites that recombine at very high levels with each other (*e.g.*, attR2 in pEZC1309 and attL2 in pEZC1321 in Table 3) as well as other recombination sites that recombine at very low levels or not at all (*e.g.*, attR1 in pEZC1305 and attLwt in pEZC1313 in Table 3). Thus, information available in the present specification and in the available art allows a ready determination of the meaning of recombination sites that do or do not "substantially recombine" with each other.

The ordinarily skilled artisan is deemed to know information that is available in the art and therefore considered well-known. *See In re Howarth*, 210 USPQ 689, 692 (CCPA 1981). Moreover, as noted above, the definiteness of claim language must always be analyzed in light of the teachings of the prior art and of the particular application disclosure as it would be interpreted by the ordinarily skilled artisan. *See Moelands*, 3 USPQ2d at 1476. Hence, whether or not two recombination sites "substantially recombine with each other" can be readily determined by one of ordinary skill in the art in view of the teachings of the present specification, and the scope of claims 14 and 19 reciting this phrase therefore could also be readily determined by the ordinarily skilled artisan. These claims thus comport with the requirements of 35 U.S.C. § 112, second paragraph, as interpreted under *Moelands* and *Moore*.

In view of the foregoing remarks, Applicants respectfully assert that claims 14 and 19 as currently presented particularly point out and distinctly claim the subject matter regarded by Applicants as the invention. Reconsideration and withdrawal of this portion of the rejection under 35 U.S.C. § 112, second paragraph, are therefore respectfully requested.

C. The Recitation of "Two or More Recombination Sites"

The Examiner next contends that claim 19 is indefinite for reciting "two or more different Vector Donor molecules comprising two or more recombination sites." *See* Office Action, paragraph bridging pages 3-4. Specifically, the Examiner contends that it is unclear whether or not each of the Vector Donor molecules contains two or more recombination sites. By the foregoing amendments, and as suggested by the Examiner, claim 19 has been amended to indicate that *each* of the Vector Donor molecules used in the method of claim 19 (and in those of claims 14 and 15 which have been similarly amended despite not being included in this rejection) contains two or more recombination sites. Therefore, this portion of the rejection has been fully accommodated; reconsideration and withdrawal are respectfully requested.

D. The Dependence of Claim 31 Upon a Nonelected Claim

The Examiner next contends that claim 31 is vague and indefinite for depending from nonelected claim 1. *See* Office Action at page 4, first full paragraph. By the foregoing amendments, claim 31 has been amended to delete the dependency from claim 1 and to incorporate the limitations of claim 1 into claim 31 (which is now an independent claim), as suggested by the Examiner. Therefore, this portion of the rejection has been fully accommodated; reconsideration and withdrawal are respectfully requested.

E. The Recitation of "Derived From"

The Examiner next contends that the use of the term "derived from" in claims 32 and 33 renders these claims indefinite. *See* Office Action at page 4, second full paragraph. By the foregoing amendments, claims 32 and 33 have been amended as suggested by the Examiner to delete "derived" and substitute therefor -- obtained --. Therefore, this portion of the rejection has been fully accommodated; reconsideration and withdrawal are respectfully requested.

F. The Recitation of "Propagation" and "Replication"

The Examiner next contends that claims 37 and 38 are vague and indefinite for reciting a vector "which propagates and/or replicates," apparently because the terms "propagate" and "replicate" may be redundant. *See* Office Action at page 4, third full paragraph. By the foregoing amendments, claims 37 and 38 have been amended to delete "propagate and/or" from each of these claims, as suggested by the Examiner. Therefore, this portion of the rejection has been fully accommodated; reconsideration and withdrawal are respectfully requested.

G. The Recitation of "And/or Insect Cells"

In the Office Action at page 4, last paragraph, the Examiner contends that claim 37 is vague and indefinite for using the term "and/or" at the end of a list of possible host cells. By the foregoing amendments, claim 37 has been amended to delete "and/or" and to insert therefor the alternative "or." However, Applicants wish to point out that the use of this term should not be construed to mean that the vectors encompassed by claim 37 as currently presented (and by claim 38 as originally presented which also uses the term "or" in the listing of possible host cells)

replicate only in *one* of the recited host cells and not in the others recited in these claims. Indeed, as one of ordinary skill would appreciate, a number of the vectors encompassed by these claims (and described in detail in the present specification) may replicate in more than one of the recited host cells. Thus, the use of the term "or" should not be construed as being exclusive in the context in which it is used in claims 37 and 38.

This portion of the rejection therefore has been fully accommodated; reconsideration and withdrawal are respectfully requested.

H. The Alleged Omission of Essential Steps in Claim 40

The Examiner next contends that claim 40 is indefinite as allegedly being incomplete for omitting essential steps. *See* Office Action at page 5, lines 1-4. By the foregoing amendments, claim 40 has been amended to recite contacting at least two nucleic acid molecules, each comprising at least one recombination site, with at least one ribosomal protein and at least one recombination protein, and incubating the resulting mixture under conditions favoring the production of at least one product molecule. These amendments to claim 40 add no new matter, and in fact have been suggested by the Examiner in the present Office Action. *See* Office Action at page 5, lines 3-4. Thus, this portion of the rejection has been fully accommodated; reconsideration and withdrawal are respectfully requested.

I. Summary

For the foregoing reasons, Applicants respectfully assert that the claims as currently presented particularly point out and distinctly claim the subject matter regarded by Applicants

as the invention. Reconsideration and withdrawal of the rejection of claims 14-51 under 35 U.S.C. § 112, second paragraph, are therefore respectfully requested.

VI. The Rejection Under 35 U.S.C. § 102(b) over Nash

In the Office Action at pages 5-6, the Examiner rejected claims 31-32, 36 and 38-51 under 35 U.S.C. § 102(b) as being anticipated by Nash, *Meth. Enzymol. 100*:210-216 (1983) (Doc. AS34, of record; hereinafter "Nash"). Applicants respectfully traverse this rejection.

In making this rejection, the Examiner contends that:

> [s]ince Int was purified [in Nash] from E.coli cells after overexpression of Int from a plasmid bearing the Int gene, it is inherent that the crude extracts used for the in vitro assay would include the E.coli ribosomal proteins, integration host factor (IHF), HU and the Int recombinase. Nash also teaches the addition of crude preparations of IHF to in vitro recombination mixtures to enhance recombination (page 215, second full paragraph).

Office Action at page 6, lines 2-6. Applicants respectfully disagree with these contentions.

Under 35 U.S.C. § 102, a claim can only be anticipated if every element in the claim is expressly or inherently disclosed in a single prior art reference. *See Kalman v. Kimberly Clark Corp.*, 713 F.2d 760, 771 (Fed. Cir. 1983), *cert. denied*, 465 U.S. 1026 (1984). Moreover, "[t]o anticipate a claim, a reference must disclose every element of the challenged claim and enable one skilled in the art to make the anticipating subject matter." *PPG Industries, Inc. v. Guardian Industries Corp.*, 75 F.3d 1558, 1566 (Fed. Cir. 1996). Neither of these requirements is met by the disclosure of Nash.

The invention as claimed is drawn to methods of recombinational cloning using compositions comprising at least one ribosomal protein. In contrast, the compositions disclosed

in Nash do not expressly contain at least one ribosomal protein. In fact, Nash contains no explicit disclosure that the compositions used therein contain at least one ribosomal protein. Thus, Nash fails to expressly disclose all of the elements of the invention as presently claimed.

Apparently recognizing that the disclosure of Nash fails to expressly anticipate the presently claimed invention, the Examiner instead contends that Nash *inherently* discloses the invention. Applicants respectfully disagree with this contention, and wish to remind the Examiner that "[i]n order for a disclosure to be inherent . . . the missing descriptive matter must *necessarily* be present in the [cited reference] such that one skilled in the art would recognize such a disclosure." *Tronzo v. Biomet, Inc.*, 156 F.3d 1154, 1159 (Fed. Cir. 1998) (emphasis added). Moreover, to rely on an inherency argument, "the examiner must provide a basis in fact and/or technical reasoning to reasonably support the determination that the allegedly inherent characteristic *necessarily* flows from the teachings of the applied prior art." *Ex parte Levy*, 17 USPQ2d 1461, 1464 (PTO Bd. Pat. App. Int. 1990) (emphasis in original). In the present case, the Examiner has pointed to no disclosure in Nash indicating that ribosomal proteins are "*necessarily* present" in the recombination reaction mixtures used in Nash (thus the *Tronzo* standard is not met by Nash). Analogously, the Examiner has pointed to no disclosure in Nash indicating that the use of ribosomal proteins in recombination reaction mixtures would "*necessarily* flow" from the disclosure of this reference (thus the *Levy* standard is not met by Nash). At best, one of ordinary skill could only assume (as the Examiner has apparently assumed) from reading Nash that it is *possible* that ribosomal proteins are present in the reaction mixtures used therein. However, inherency "may not be established by probabilities or possibilities. The mere fact that a certain thing *may* result from a given set of circumstances is

not sufficient." *Continental Can Co. USA, Inc. v. Monsanto Co.*, 948 F.2d 1264, 1269 (Fed. Cir. 1991) (emphasis added). As one of ordinary skill would understand, there are many ways by which a crude cell extract could be prepared for use in a recombination reaction mixture that might exclude ribosomal proteins from the mixture. Therefore, although it may be *possible* that ribosomal proteins were present in the mixtures used in Nash, there is no objective indication that ribosomal proteins were *definitely* present therein (thus, the *Continental Can* standard is not met by Nash). Hence, the Examiner's attempted reliance upon inherent anticipation in the present case is factually and legally unfounded.

In view of the foregoing remarks, and under 35 U.S.C. § 102(b) in view of *Kalman* and *PPG Industries*, Applicants respectfully assert that Nash cannot and does not anticipate the invention as presently claimed. Reconsideration and withdrawal of the rejection of claims 31-32, 36 and 38-51 under 35 U.S.C. § 102(b) are therefore respectfully requested.

VII. The Rejection Under 35 U.S.C. § 102(b) Over Abremski I and Abremski II

In the Office Action at page 6, the Examiner has rejected claims 40-51 under 35 U.S.C. § 102(b) as being anticipated by Abremski *et al.*, *J. Biol. Chem.* 259:1509-1514 (1984) (Doc. AS1, of record; hereinafter "Abremski I") and Abremski *et al.*, *J. Biol. Chem.* 257:9658-9662 (Doc. AR1, of record; hereinafter "Abremski II"). Applicants respectfully traverse this rejection.

In making this rejection, the Examiner contends that:

> [i]n both [Abremski I and Abremski II], the enzymes were prepared from crude extracts of E.coli cells in which the enzymes were overexpressed and the enzymatic activity followed throughout the purification process (Table I of both papers). It is reasonable to expect that the E.coli ribosomal proteins as well [as]

> the E.coli proteins IHF and HU would have been present in each of the crude extracts tested.

Office Action at page 6, second full paragraph, lines 5-9. Applicants respectfully disagree with these contentions.

The invention as claimed in claim 40 (and thus the remaining claims depending therefrom) is drawn to methods of enhancement of recombinational cloning by contacting a nucleic acid molecule with one or more ribosomal proteins and one or more recombination proteins under conditions favoring the enhancement of cloning of the nucleic acid molecule. In contrast, the compositions disclosed in Abremski I and Abremski II do not expressly contain at least one ribosomal protein. In fact, these papers provide no explicit disclosure that the compositions used therein contain at least one ribosomal protein. Thus, Abremski I and Abremski II fail to expressly disclose all of the elements of the invention as presently claimed.

Again apparently recognizing the insufficiency of the disclosure of these papers, the Examiner relies instead on inherency to support this rejection. Applicants wish to remind the Examiner, however, that it is irrelevant whether or not it is "reasonable" to expect that ribosomal proteins would be present in crude cell extracts, since as noted above under *Tronzo* and *Levy*, one of ordinary skill would have to conclude that ribosomal proteins are *necessarily* (not *reasonably*) present in the compositions used in the cited references. Since the Examiner has not pointed to any disclosure in Abremski I or Abremski II indicating that ribosomal proteins are *necessarily* present in the reaction mixtures used therein, the Examiner's attempted reliance upon inherent anticipation in the present case is factually and legally unfounded. Thus, Applicants respectfully assert that the invention as presently claimed is neither expressly nor inherently disclosed in Abremski I and/or Abremski II.

In view of the foregoing remarks, and under 35 U.S.C. § 102(b) in view of *Kalman* and *PPG Industries*, Applicants respectfully assert that Abremski I and Abremski II cannot and do not anticipate the invention as presently claimed. Reconsideration and withdrawal of the rejection of claims 40-51 under 35 U.S.C. § 102(b) are therefore respectfully requested.

VIII. The Rejection Under 35 U.S.C. § 102(e) Over Hartley

In the Office Action at page 7, the Examiner has rejected claims 14-51 under 35 U.S.C. § 102(e) as being anticipated by Hartley. Applicants respectfully traverse this rejection.

In making this rejection, the Examiner contends that:

> [i]n vitro applications would be expected to encompass embodiments wherein the methods are practiced with crude E.coli lysates comprising one or more recombination factors (e.g. IHF, HU and a recombinantly expressed recombinase) and wherein one or more E.coli ribosomal proteins would be present.

Office Action at page 7, second paragraph, lines 3-6. Applicants respectfully disagree with these contentions.

As noted above, the invention as claimed is drawn to methods of recombinational cloning using compositions comprising at least one ribosomal protein. In contrast, the compositions disclosed in Hartley do not expressly contain at least one ribosomal protein. In fact, this cited reference provides no explicit disclosure that the compositions used therein contain at least one ribosomal protein. Thus, Hartley fails to expressly disclose all of the elements of the invention as presently claimed.

Again apparently recognizing the insufficiency of the disclosure of Hartley, the Examiner relies instead on inherency to support this rejection. Applicants again note that under *Tronzo* and

Levy, one of ordinary skill would have to conclude that ribosomal proteins are *necessarily* present in the compositions used in Hartley in order for this reference to inherently anticipate the presently claimed invention. Since the Examiner has not pointed to any disclosure in Hartley indicating that ribosomal proteins are *necessarily* present in the reaction mixtures used therein, the Examiner's attempted reliance upon inherent anticipation in the present case is factually and legally unfounded. In fact, the Examiner appears to acknowledge this fact by stating, in two locations in the Office Action, that:

> it could be considered that in vitro embodiments of the methods taught by Hartley et al comprising the use of a crude extract from E.coli having one or more recombination factors and ribosomal proteins for recombinational cloning is not encompassed by the teachings of [Hartley] . . . .
>
> * * * *
>
> Hartley et al do not explicitly teach the use of crude lysates comprising recombination factors in their in vitro methods. Hartley et al do not explicitly teach the addition of ribosomal proteins to their recombination mixtures.

Office Action at page 7, third paragraph, lines 1-4, and at page 8, third full paragraph, respectively. Thus, Applicants respectfully assert that the invention as presently claimed is neither expressly nor inherently disclosed in Hartley.

In view of the foregoing remarks, and under 35 U.S.C. § 102(e) in view of *Kalman* and *PPG Industries*, Applicants respectfully assert that Hartley cannot and does not anticipate the invention as presently claimed. Reconsideration and withdrawal of the rejection of claims 14-51 under 35 U.S.C. § 102(e) are therefore respectfully requested.

IX. The Rejection Under 35 U.S.C. § 103(a)

In the Office Action at pages 7-9, the Examiner has rejected claims 14-51 under 35 U.S.C. § 103(a) as being unpatentable over Hartley in view of Nash or Abremski I or Abremski II. Applicants respectfully traverse this rejection.

In proceedings before the Patent and Trademark Office, the examiner bears the burden of establishing a *prima facie* case of obviousness based upon the prior art. *See In re Piasecki*, 223 USPQ 785, 787-88 (Fed. Cir. 1984). The Examiner can satisfy this burden only by showing some objective teaching in the prior art or that knowledge generally available to one of ordinary skill in the art would lead that individual to combine the relevant teachings of the references in such a way as to produce the invention as claimed, *see In re Fine*, 5 USPQ2d 1596,1598 (Fed. Cir: 1988), and that would also suggest a reasonable likelihood of success in making or using the claimed invention as a result of that combination. *See In re Dow Chem. Co.*, 837 F.2d 469, 473 (Fed. Cir. 1988). In the present case, the Examiner's burden has not been satisfied.

As discussed above, Hartley does not disclose or suggest the presently claimed invention for the reasons detailed above which are reiterated and incorporated herein by reference. As also discussed above, while acknowledging that Hartley does not teach every limitation of the claimed invention that would make this reference relevant as a primary reference for use in an obviousness rejection, the Examiner appears to view the information that is explicitly missing from Hartley as inherent in that reference, or at least as within the level of ordinary skill in the art. Applicants wish to remind the Examiner, however, that there is no such thing as "inherent obviousness," since inherence and obviousness are different legal concepts. *See In re Spormann*, 150 USPQ 449, 452 (C.C.P.A. 1966). That which is inherent cannot be obvious, since inherent

information "is not necessarily known . . . . [and] Obviousness cannot be predicated on what is unknown." *Id.* Since the present rejection is based on obviousness, any contention by the Examiner that is based on the possible presence of inherent knowledge in Hartley must necessarily fail. Hence, Hartley is seriously deficient as a primary reference upon which to base a rejection under 35 U.S.C. § 103(a).

These deficiencies in Hartley are not cured by the disclosures of Nash, Abremski I or Abremski II. As noted above, none of these references provide explicit disclosure of the use of ribosomal proteins in recombination reaction mixtures. Indeed, even if these references *inherently* disclosed the use of ribosomal proteins (which, as discussed above, is not the case), inherency cannot form the basis of an obviousness rejection under *Spormann*. Finally, there is no disclosure, suggestion, or contemplation in any of these references that would have motivated one of ordinary skill to combine their disclosures so as to produce the presently claimed methods with any reasonable expectation of success, since none of the secondary references discloses recombinational cloning methods using reaction mixtures that *necessarily* contain one or more ribosomal proteins. Absent such suggestion, motivation and reasonable expectation of success, the cited references may not be properly combined to render the claimed invention obvious. *See Fine*, 5 USPQ2d at1598; *see also Dow Chem. Co.*, 837 F.2d at 473.

Thus, the statutory burden required to sustain a *prima facie* case of obviousness has not been met. Reconsideration and withdrawal of the rejection under 35 U.S.C. § 103(a) over Hartley in view of Nash, Abremski I and Abremski II therefore are respectfully requested.

X. *The Double Patenting Rejection*

In the Office Action at pages 9-10, the Examiner has rejected claims 14-51 under the judicially created doctrine of obviousness-type double patenting as being unpatentable over claims 29-37 of Hartley. Applicants respectfully traverse this rejection, and respectfully disagree with the Examiner's contention that claims 14-51 in the present application are not patentably distinct from claims 29-37 of Hartley for the reasons noted above concerning the disclosure of Hartley, which are reiterated and incorporated by reference herein. However, Applicants respectfully request that this rejection be held in abeyance until identification of patentable subject matter in the present application.

XI. *Other Matters*

Applicants acknowledge receipt of the Examiner-initialed copy of the Form PTO-1449 submitted with Applicants' Information Disclosure Statement (IDS) filed on September 18, 2000, in the present matter. Applicants also acknowledge the Examiner's comments in the Office Action at page 2, second paragraph, that copies of the references cited on the Form PTO-1449 were not located with the file. Applicants thank the Examiner for the time taken to locate a number of these cited references in a prior application and to consider them in connection with the present application. Enclosed herewith are copies of the return receipt postcard bearing the PTO date stamp of September 18, 2000, indicating receipt by the U.S. Patent and Trademark Office of copies of the 234 references cited in connection with the above-referenced IDS. Also enclosed herewith are courtesy copies of the following documents cited on the Form PTO-1449 attached to the above-referenced IDS, which the Examiner has indicated as not being located

with the present file: AT1, AS4, AT4, AR5, AR9, AR19, AT21, AS22, AS24, AS26, AS29, AR32, AS32, AR35, AT36, AS38, AT40, AR46, AS46, AR47, AR49, AS49, AR50, AS52, AR53, AR54, AR57, AT58 and AS59-AS62. It is respectfully requested that the Examiner initial and return an additional or revised copy of the Form PTO-1449 that was filed in the present matter on September 18, 2000, and indicate in the official file wrapper of this patent application that these specific documents have been considered.

XII. Conclusion

All of the stated grounds of rejection have been properly traversed, accommodated, or rendered moot. Applicants therefore respectfully request that the Examiner reconsider all presently outstanding rejections and that they be withdrawn.

Applicants believe that a full and complete reply has been made to the outstanding Office Action and, as such, the present application is in condition for allowance. If the Examiner believes, for any reason, that personal communication will expedite prosecution of this application, the Examiner is invited to telephone the undersigned at the number provided.

Respectfully submitted,

STERNE, KESSLER, GOLDSTEIN & FOX P.L.L.C.

Brian J. Del Buono
Attorney for Applicants
Registration No. 42,473

Date: March 8 2001

1100 New York Avenue, N.W.
Suite 600
Washington, D.C. 20005-3934
(202) 371-2600
P100-69.wpd

Version with markings to show changes made

In the Specification:

In the specification at page 51, the paragraph appearing at lines 21-29 has been amended as follows:

-- Plasmid pEZ13835 (Figure 6; *attP*), pEZC7501 (Figure 7; *attB*), pEZ11104 (Figure 8; *attR*), and pEZC8402 (Figure 9; *attL*) were as shown. pEZC7501 was cut with *Sca*I and pEZC8402 with *Nco*I before use. pEZ13835 and pEZC8402 were propagated in *E. coli* DB2 and the other two in *E. coli* DH5α. Cells from a glycerol seed were placed in 25 ml of [Circlegrow] CIRCLEGROW® brand culture medium (BIO 101) plus 100 mg/ml ampicillin (pEZC7501 and pEZC8402) or plus 100 mg/ml kanamycin (pEZ13835 and pEZ11104) and grown overnight at 37 °C. Cells were harvested by centrifugation and stored at - 70 °C. Plasmid DNAs were purified using Qiagen Midi products and protocols. --

In the specification at pages 62-63, the paragraph appearing at page 62, line 27, through page 63, line 4, has been amended as follows:

-- *Growth of Cells.* Cells from a glycerol stock of BL21DE3 bearing plasmid pET12AS20AA were inoculated into 3 ml of LB broth containing 100 mg/ml ampicillin. This inoculum was diluted into LB broth + 100 mg/ml ampicillin 1:100 and the 300-ml culture was grown overnight at 30 °C. The A_{650} of the culture should not exceed 1.0. This culture was used to innoculate 10 flasks containing 500 ml each of CIRCLEGROW® brand culture medium

(BIO 101) plus 100 mg/ml ampicillin plus 1 mM $MgSO_4$. Cells were grown at 37 °C until the A_{650} was 0.5 and expression of S20 was induced by the addition of IPTG to 0.5 mM. After growth at 37 °C for 4 hours, cells were harvested by centrifugation at 4 °C and stored at -70 °C. --

In the Claims:

The claims have been amended as follows:

(a) Claims 1-13 and 52-64 have been cancelled, without prejudice to or disclaimer of the subject matter encompassed thereby.

(b) Currently pending claims 14, 15, 19, 29, 31-33, 37-40 and 50 have been amended as follows:

14. (Once amended) A method for cloning or subcloning one or more desired nucleic acid molecules comprising

(a) forming a combination by combining *in vitro* [or *in vivo*]

(i) one or more Insert Donor molecules comprising one or more desired nucleic acid segments flanked by at least two recombination sites, wherein said recombination sites do not substantially recombine with each other;

(ii) one or more Vector Donor molecules <u>each</u> comprising at least two recombination sites, wherein said recombination sites do not substantially recombine with each other;

(iii) an effective amount of at least one recombination protein; and

(iv) an effective amount of at least one ribosomal protein; and

(b) incubating said combination under conditions sufficient to transfer one or more of said desired segments into one or more of said Vector Donor molecules, thereby producing one or more desired Product nucleic acid molecules.

15. (Once amended) The method of claim 14, further comprising:

(c) forming a combination by combining *in vitro* [or *in vivo*]

(i) one or more of said Product molecules comprising said desired segments flanked by two or more recombination sites, wherein said recombination sites do not substantially recombine with each other;

(ii) one or more different Vector Donor molecules <u>each</u> comprising two or more recombination sites, wherein said recombination sites do not substantially recombine with each other;

(iii) an effective amount of at least one recombination protein; and

(iv) an effective amount of at least one ribosomal protein; and

(d) incubating said combination under conditions sufficient to transfer one or more of said desired segments into one or more different Vector Donor molecules, thereby producing one or more different Product molecules.

19. (Once amended) A method for cloning or subcloning desired nucleic acid molecules comprising

a) forming a combination by combining *in vitro* [or *in vivo*]

i) one or more Insert Donor molecules comprising one or more nucleic acid segments flanked by two or more recombination sites, wherein said recombination sites do not substantially recombine with each other;

ii) two or more different Vector Donor molecules each comprising two or more recombination sites, wherein said recombination sites do not substantially recombine with each other;

iii) an effective amount of at least one recombination protein; and

iv) an effective amount of at least one ribosomal protein; and

b) incubating said combination under conditions sufficient to transfer one or more of said desired segments into said different Vector Donor molecules, thereby producing two or more different Product molecules.

29. (Once amended) The method of claim 14, wherein said recombination protein is selected from the group consisting of Int, Cre, FLP, Xis, IHF, FIS and HU, and combinations thereof.

31. (Once amended) A method for recombinational cloning of one or more desired nucleic acid molecules comprising

(a) forming a mixture by mixing *in vitro* one or more of said desired nucleic acid molecules with one or more vectors and with [the composition of claim 1; and] an effective

amount of at least one ribosomal protein and an effective amount of at least one recombination protein; and

(b) incubating said mixture under conditions sufficient to transfer said one or more desired nucleic acid molecules into one or more of said vectors.

32. (Once amended) The method of claim 31, wherein said desired nucleic acid molecules are [derived] obtained from genomic DNA.

33. (Once amended) The method of claim 31, wherein said desired nucleic acid molecules are [derived] obtained from cDNA.

37. (Once amended) The method of claim 36, wherein said eukaryotic vector [propagates and/or] replicates in yeast cells, plant cells, fish cells, eukaryotic cells, mammalian cells, [and/or] or insect cells.

38. (Once amended) The method of claim 31, wherein said prokaryotic vector [propagates and/or] replicates in bacteria of the genera *Escherichia, Salmonella, Bacillus, Streptomyces* or *Pseudomonas*.

39. (Once amended) The method of claim 38, wherein said prokaryotic vector [propagates and/or] replicates in *E. coli*.

40. (Once amended) A method for enhancement of recombinational cloning, comprising contacting [a nucleic acid molecule] at least two nucleic acid molecules each comprising at least one recombination site *in vitro* with one or more ribosomal proteins and with one or more recombination [proteins.] proteins to form a mixture, and incubating said mixture under conditions favoring the production of at least one product nucleic acid molecule.

50. (Once amended) The method of claim 40, wherein said recombination protein is selected from the group consisting of Int, Cre, FLP, Xis, IHF, FIS and HU, and combinations thereof.

(c) New claims 65-88 are sought to be added.

information "is not necessarily known . . . . [and] Obviousness cannot be predicated on what is unknown." *Id.* Since the present rejection is based on obviousness, any contention by the Examiner that is based on the possible presence of inherent knowledge in Hartley must necessarily fail. Hence, Hartley is seriously deficient as a primary reference upon which to base a rejection under 35 U.S.C. § 103(a).

These deficiencies in Hartley are not cured by the disclosures of Nash, Abremski I or Abremski II. As noted above, none of these references provide explicit disclosure of the use of ribosomal proteins in recombination reaction mixtures. Indeed, even if these references *inherently* disclosed the use of ribosomal proteins (which, as discussed above, is not the case), inherency cannot form the basis of an obviousness rejection under *Spormann*. Finally, there is no disclosure, suggestion, or contemplation in any of these references that would have motivated one of ordinary skill to combine their disclosures so as to produce the presently claimed methods with any reasonable expectation of success, since none of the secondary references discloses recombinational cloning methods using reaction mixtures that *necessarily* contain one or more ribosomal proteins. Absent such suggestion, motivation and reasonable expectation of success, the cited references may not be properly combined to render the claimed invention obvious. *See Fine*, 5 USPQ2d at1598; *see also Dow Chem. Co.*, 837 F.2d at 473.

Thus, the statutory burden required to sustain a *prima facie* case of obviousness has not been met. Reconsideration and withdrawal of the rejection under 35 U.S.C. § 103(a) over Hartley in view of Nash, Abremski I and Abremski II therefore are respectfully requested.